High concentrations of dipeptidyl peptidase-4 inhibitors suppressed lipopolysaccharide-induced inflammatory cytokine expression in U937 cells

Shiho Yamadera¹, Yuya Nakamura², Masahiro Inagaki³, Isao Ohsawa⁴, Yoshikazu Goto⁴, Hiromichi Gotoh⁴, Mayumi Tsuji², Shinichi Iwai¹, Yuji Kiuchi²

¹Department of Healthcare and Regulatory Sciences, Showa University School of Pharmacy, Tokyo, Japan;
²Department of Pharmacology, Showa University School of Medicine, Tokyo, Japan;
³Department of Chemistry, College of Arts and Sciences, Showa University, Tokyo, Japan;
⁴Saiyu Soka Hospital, Saitama, Japan

Background
Dipeptidyl peptidase-4 (DPP-4) inhibitors known as enhancers of incretin hormones such as glucagon-like peptide-1 are therapeutic drugs used in patients with type II diabetes mellitus. Furthermore, DPP-4 inhibitors have been indicated to have the anti-inflammatory potential in various inflammatory models. However, few studies have investigated the anti-inflammatory properties of DPP-4 inhibitors using monocyteic cells that play an important role in atherosclerosis-associated inflammation. In the present study, we examined the anti-inflammatory effect of DPP-4 inhibitors on lipopolysaccharide (LPS)-induced inflammation in human monocytic cell line U937.

Methods
U937 cells (1 × 10⁶ cells/mL) were cultured in RPMI-1640 medium supplemented with 10% fetal bovine serum and 1% antibiotic-antimycotic solution, at 37 °C, with 5% CO₂. The cells were differentiated into macrophages by incubation of phorbol 12-myristate 13-acetate at 100 nM/mL for 48 hrs, and subsequently the differentiated cells were cultured with or without 1ng/mL LPS (used as a control) for 5 hrs at 37 °C. To study the suppressive effects of DPP-4 inhibitors against LPS-induced inflammation, we pretreated U937 cells with Linagliptin (100, 250, 500 and 2,500 nM) or Sitagliptin (250, 500, 1,000 and 5,000 nM) for 1 hr followed by treatment with LPS (1 ng/mL) + DPP4 inhibitors (the concentration used for pretreatment) for 5 hrs. To evaluate the inflammation, interleukin (IL)-6 levels in cell culture supernatant were measured using a human IL-6 ELISA kit after incubation for 5 hrs.

Results
The levels of IL-6 in the LPS-exposed cells were significantly increased, which were prevented by the pretreatment with Linagliptin or Sitagliptin in a dose-dependent manner. Especially, pretreated cells with high concentrations of Linagliptin (500 nM and 2,500 nM) or Sitagliptin (1,000 nM and 5,000 nM) significantly reduced IL-6 levels increased by LPS exposure. However, treated cells with both DPP-4 inhibitors had no significant effect compared with the control cells.

Conclusions
In this study, we indicated that high concentrations of Linagliptin or Sitagliptin which are therapeutic agents in the treatment of type II diabetes mellitus might suppress LPS-induced inflammation in human macrophage U937 cells through a strong attenuation of IL-6 expression.